Amendments to the Claims

This listing of claims will replace all prior versions, and listings, of claims in the application.

Listing of claims:

Claims 1-89. (Cancelled)

90. (Currently Amended) A method of generating a frozen viable cartilage, comprising:

(a) providing a receptacle containing [[a]] viable cartilage in a cryopreservation solution at a temperature above a freezing temperature of the cryopreservation solution;

(b) cooling the viable cartilage in the cryopreservation solution to a temperature below the freezing temperature of the cryopreservation solution at a cooling rate of 0.01°C/min to 3°C/min, thereby generating a frozen viable cartilage in the receptacle; and

(c) transferring the receptacle to storage at a temperature equal to or below -130°C,

~~wherein upon thawing the thawed viable cartilage comprises more than 50% viable chondrocytes~~.

91. (Previously Presented) The method of claim 90, wherein cooling comprises moving the receptacle along one or more consecutive temperature gradients ranging from a temperature above the freezing temperature to a temperature below the freezing temperature.

92. (Currently Amended) The method of claim 90, wherein cooling comprises ~~controlled initiation of~~ controllably initiating seeding ~~of freezing~~.

93. (Previously Presented) The method of claim 90, wherein the viable cartilage comprises osteochondral tissue.

94. (Currently Amended) The method of claim 90 [[91]], wherein movement along the at least one temperature gradient is at a velocity between 0.002 mm/sec and 5 mm/sec and at a cooling rate of between 0.1°C/mm to 50°C/mm.

95. (Previously Presented) The method of claim 91, wherein at least one of the one or more consecutive temperature gradients is between 0.1°C/mm to 50°C/mm.

96. (Previously Presented) Frozen viable cartilage produced by the method of claim 91.

97. (Currently Amended) A method for thawing a frozen viable cartilage that was frozen in a cryopreservation-solution, the method comprising:

(a) providing a receptacle containing [[the]] frozen viable cartilage at an initial temperature below the glass transition temperature of the cryopreservation-solution;

(b) warming the receptacle containing the frozen viable cartilage and the cryopreservation solution from the initial temperature to an intermediate temperature being ~~at least~~ about the glass transition temperature or above the glass transition temperature of the cryopreservation solution but no more than the transition temperature of the cryopreservation solution wherein recrystalization would begin to occur at any point in the cartilage; and

(c) warming the frozen viable cartilage and the cryopreservation solution from the intermediate temperature to a temperature that is at least substantially equal to the melting temperature of the cryopreservation solution, the warming being at a rate sufficiently high to minimize recrystalization to obtain ~~; thereby obtaining~~ thawed viable cartilage~~,~~

~~wherein upon thawing the thawed viable cartilage comprises more than 50% viable chondrocytes~~.

98. (Currently Amended) The method of claim 97, wherein warming the receptacle containing the frozen viable cartilage and the cryopreservation solution from the initial temperature to the intermediate temperature is at a rate sufficiently slow to minimize fracture of the frozen viable cartilage.

99. (Currently Amended) The method of claim 97, wherein warming the receptacle containing the frozen viable cartilage and the cryopreservation solution from the initial temperature to the intermediate temperature is at a rate of between 0.1°C/min and 200°C/min.

100. (Previously Presented) (Previously Presented) The method of claim 99, wherein warming is at a rate of 90ºC/min.

101. (Previously Presented) The method of claim 97, wherein warming is at a rate of between 50°C/min and 1000°C/min.

102. (Previously Presented) The method of claim 101, wherein warming is at a rate of 200ºC/min.

103. (Previously Presented) The method of claim 97, wherein the intermediate temperature is less than -10ºC, or -20 to -80ºC, or -40 to -80ºC, or -50 to -70ºC.

104. (Previously Presented) The method of claim 97, wherein warming comprises:

(i) removing the frozen viable cartilage from the receptacle; and

(ii) contacting the frozen viable cartilage with an environment having the temperature that is at least substantially equal to the melting temperature of the cryopreservation solution, the temperature being 0ºC or more.

105. (Previously Presented) The method of claim 104, wherein the temperature of the environment is at least 22ºC, at least 37ºC, at least 50ºC, or at least 70ºC.

106. (Previously Presented) The method of claim 104, wherein the frozen viable cartilage is connected to a pulling member and removing comprises pulling the pulling member.

107. (Previously Presented) Thawed viable cartilage produced by the method of claim 97.

108. (Previously Presented) A method of providing a patient having impaired cartilage in an organ at a target site, with a thawed viable cartilage, the method comprising:

(a) providing the thawed viable cartilage of claim 107 having a shape and size compatible with the target site in the organ; and

(b) grafting the thawed viable cartilage in the target site,

wherein the grafted cartilage is viable.

109. (Previously Presented) The method of claim 108, wherein the organ is a joint.

110. (Previously Presented) The method of claim 108, wherein the thawed viable cartilage comprises osteochondral tissue.

111. (Previously Presented) The method of claim 90, wherein said storage is at a temperature between -130°C and -196°C.

112. (New) A method, comprising:

providing a receptacle containing frozen viable cartilage at an initial temperature below the glass transition temperature of the cryopreservation solution;

warming the receptacle containing the frozen viable cartilage and the cryopreservation solution from the initial temperature to an intermediate temperature being about the glass transition temperature or above the glass transition temperature of the cryopreservation solution but no more than the transition temperature of the cryopreservation solution wherein recrystalization would begin to occur at any point in the cartilage;

warming the frozen viable cartilage and the cryopreservation solution from the intermediate temperature to a temperature that is at least substantially equal to the melting temperature of the cryopreservation solution, the warming being at a rate sufficiently high to minimize recrystalization to obtain thawed viable cartilage;

placing the thawed viable cartilage in a buffered physiological solution comprising a non-cell membrane permeable biocompatible sugar, polyol or other organic solute.

113. (New) A method, comprising:

providing a receptacle containing viable cartilage in a cryopreservation solution at a temperature above a freezing temperature of the cryopreservation solution;

cooling the viable cartilage in the cryopreservation solution to a temperature below the freezing temperature of the cryopreservation solution at a cooling rate of 0.01°C/min to 3°C/min, thereby generating a frozen viable cartilage in the receptacle; and

transferring the receptacle to storage at a temperature equal to or below -130°C;

thawing the viable cartilage to obtain thawed viable cartilage

placing the thawed viable cartilage in a buffered physiological solution comprising a non-cell membrane permeable biocompatible sugar, polyol or other organic solute, a serum of any species and antibiotics.

114. (New) The method according to claim 112, wherein the buffered physiological solution further comprises a serum of any species.

115. (New) The method according to claim 113, wherein the buffered physiological solution further comprises a serum of any species.

116. (New) The method according to claim 112, wherein the buffered physiological solution further comprises an antibiotic.

117. (New) The method according to claim 113, wherein the buffered physiological solution further comprises an antibiotic.

118. (New) The method of claim 112, wherein the non-cell membrane permeable biocompatible sugar, polyol or other organic solute is selected from the group consisting of sucrose, mannitol, sorbitol, trehalose, fructose, glucose, raffinose, maltose, xylitol and amino acids.

119. (New) The method of claim 113, wherein the non-cell membrane permeable biocompatible sugar, polyol or other organic solute is selected from the group consisting of sucrose, mannitol, sorbitol, trehalose, fructose, glucose, raffinose, maltose, xylitol and amino acids.

120. (New) The method of claim 90, wherein upon thawing the thawed viable cartilage comprises more than 65% viable chondrocytes.

121. (New) The method of claim 97, wherein upon thawing the thawed viable cartilage comprises more than 65% viable chondrocytes.

122. (New) The method of claim 112, wherein upon thawing the thawed viable cartilage comprises more than 65% viable chondrocytes.

123. (New) The method of claim 113, wherein upon thawing the thawed viable cartilage comprises more than 65% viable chondrocytes.

124. (New) The method according to claim 90, wherein the receptacle further contains bone.

125. (New) The method according to claim 97, wherein the receptacle further contains bone.

126. (New) The method according to claim 112, wherein the receptacle further contains bone.

127. (New) The method according to claim 113, wherein the receptacle further contains bone.